especially prominent within a ~100 kbp region around a centromere where the density effects statistically stretch the chromatin. We also found that a whole region of parameters describing the average state of the chromatin fiber was consistent with the experimental Hi-C data. Finally, the dynamical simulations showed that rapid progression through cell cycle allowed for spatial, but not necessarily topological, equilibration of yeast chromosomes, limiting their mutual entanglement.

2997-Pos Board B152
Interactions of Nuclear Actin-Related Proteins with SWI/SNF Chromatin-Remodeling Complexes
Bengi Turegun, David J. Kast, Roberto Dominguez.
University of Pennsylvania, Philadelphia, PA, USA.
ATP-dependent chromatin-remodeling complexes (remodelers) modulate chromatin structure and thus play important roles in transcription, DNA replication, and DNA repair. Nuclear actin and actin-related proteins (Arps) are subunits of several such complexes, where they are commonly found in pairs. However, the role of actin/Arps and the mechanism through which they bind the catalytic subunits of their host complexes are unknown. Here, we investigated how budding yeast Arp7 and Arp9 are incorporated into SWI/SNF-family chromatin-remodeling complexes. We present the first biochemical evidence that Arp7 and Arp9 must heterodimerize to bind ATP and interact with the catalytic subunits of the SWI/SNF and RSC complexes. In addition, we present ITC data, which define the remodeler region that interacts with Arp7/9, thus providing a mechanistic framework for understanding how actin/Arps are selectively loaded into their host remodelers.

2998-Pos Board B153
Study of Nuclear Organization through the Dynamic Properties of Chromatin
Irena Bronshtein, Eldad Kepten, Moshe Lindner, Yaron Shav Tal, Yuval Garini.
Bar Ilan, Ramat Gan, Israel.
Chromosomes occupy specific nuclear volumes called chromosome territories and their motion is highly constrained. Little is known about which proteins and structures organize chromosome territories. A major object of our research is to understand the biophysical mechanisms that maintain this organization. We turned to study the diffusion properties of genome in order to shed light on this maintenance mechanism. The diffusion character of species depends on its properties and on the environment, thereby providing an excellent method for studying the nuclear maintenance mechanism. We examined genome mobility by focusing on three different genomic elements: telomeres, centromeres and specific gene loci. We developed method that allows measuring the diffusion in time-range of 10^{-2} - 10^{4} sec. Such broad time range allowed us to identify the transient anomalous diffusion of different genomic regions that could not be identified by other techniques. Anomalous diffusion usually depends on environmental constrains, such as temporal binding. Therefore, we propose a model for chromatin organization maintenance in the nucleus that is based on temporal binding of chromatin to itself, or to other nuclear entities. In order to prove this hypothesis, we decided to focus on identifying the possible molecular source of the suggested binding. We conduct our research on measuring the effect of loss of Lamin A on chromatin's diffusion properties. We found that telomeres and centromes motion in cells without Lamin A is ~8 times less constrained compared to normal cells. It also shows normal diffusion, while in normal cells diffusion was found anomalous. Based on our results we can conclude that lack of Lamin A leads to looser chromatin. Finding other proteins that are responsible for such binding is a great challenge that we are now pursuing.

2999-Pos Board B154
Exploring the Chromatin Architecture in Living Cells by Minutes-Long Tracking of Gold Nanoparticles
Laura C. Estrada[1], Enrico Gratton[2].
[1]University of Buenos Aires and IFIBA, Buenos Aires, Argentina, [2]Laboratory for Fluorescence Dynamics, Department of Biomedical Engineering, University of California, Irvine, USA, irvine, CA, USA.
Investigating the chromatin compaction on the micro(nano)-meter scale, has become a question of interest to understand many cellular processes. Previous evidence suggests that the cell nucleus is spatially heterogeneous and with inaccessible regions mainly due to a high concentration of chromatin. However, the in vivo 3D picture of the nuclear structure remains unclear. In this work, we studied chromatin organization applying the orbital 3D tracking technique to 20 nm gold nanoparticles (NPs) previously incorporated inside the nucleus of NIH3T3 live cells. We have recently shown that metallic NPs do not bleach or blink upon continuous illumination, are extremely stable, very bright and their luminescence spans over the visible spectrum. These characteristics allow us to track them for minutes thus providing 3D trajectories appreciably longer than those based on fluorescent proteins or quantum dots. For this study we have analyzed the motion of 60 NPs. Each one provided us with a ~5 - 30 minutes long trajectory. In ~30% of the cases, we have observed that the NPs remain in regions of apparent confined motion (clusters) and eventually they undergo a long (in the micrometer range) excursion. We have found that the NPs always move faster within the clusters but slower while travelling between two clusters. These results suggest that the NPs get trapped into cavities where they can move relatively fast and eventually get transported (as seen by MSD analysis) from one cavity to the next one along segments of slower diffusion. Additionally, in all the cases analyzed, the NPs showed an increased intensity while moving between two cavities.
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3000-Pos Board B155
Distinct Dosage Compensation Effects by Subunits of Drosophila MSL Complexes
David D. Dunlap[1], Ruth Yokoyama[2], Huiping Ling[2], He-Ying Sun[2], Kerry McGill[1], Simona Cugusi[2], John Lucchesi[2].
[1]Emory University School of Medicine, Atlanta, GA, USA, [2]Emory University, Atlanta, GA, USA.
Epigenetic regulation at the chromosomal level compensates for the difference in the dosage of X-linked genes between the sexes (dosage compensation). In Drosophila, this regulatory mechanism operates through the MSL complex that enhances the transcription of many genes on the X chromosome in males. The results of an investigation of the structural effects of various subunits of the complex confirmed that enriched, specific acetylation of histone H4 at lysine 16 by the histone acetyl transferase subunit MOF induced a more disorganized state of reconstituted single chromatin fibers. In addition, targeting of the MSL complex to plasmids by inclusion of the MSL assembly locus reduced the level of negative supercoiling. Similar targeting of incomplete complexes distinguished the roles that the various subunits of the complex play in this topological modification. Finally, the potential contribution of ISWI containing remodeling complexes to the architecture of compensated chromatin was analyzed and the results indicate a probable role for this remodeling factor in dosage compensation.

3001-Pos Board B156
Rabl Organization of Chromosomes in the Yeast Nucleus
Gabriel Bronk, Baris Avsaroglu, Jungoh Ham, James Haber, Jane Kondev.
Brandeis University, Waltham, MA, USA.
The location of genes in the interphase nucleus can influence gene expression and recombination. We examine a random walk polymer model of an interphase yeast chromosome that takes into account Rabl organization, namely attachment of the centromere to the spindle pole body, and tethering of the telomeres to the nuclear membrane. using this model, we calculate the probability distribution for the spatial positioning of a single genetic locus on chromosome III and compare it to an experimental distribution obtained by fluorescence microscopy of wild-type yeast cells. To best fit the model to the experimental distribution, the parameters for chromatin rigidity and nuclear architecture are optimized using values within the ranges reported by previous studies. We then quantitatively test the model using a yeast mutant in which the telomeres are not tethered to the nuclear envelope. The mutant's experimental and computational distributions quantitatively agree, which is evidence that a random walk polymer model of yeast chromosomes that incorporates Rabl organization can account for the spatial positioning of genetic loci during interphase. Further studies will apply this model to the understanding of homologous recombination, specifically in the context of double strand break repair.

3002-Pos Board B157
Single-Molecule Studies of a ParB Family Chromosome Segregation Protein from *Bacillussubtilis*
Thomas G.W. Graham[1], **Linda Song**[1], Xindan Wang[1], Candice M. Etson[2], Antoine Van Oijen[3], David Z. Rudner[1], Joseph J. Loparo[1].
[1]Harvard Medical School, Boston, MA, USA, [2]Tufts University, Medford, MA, USA, [3]Zernike Institute for Advanced Materials, Rijksuniversiteit Groningen, Groningen, Netherlands.
ParAB systems play a role in chromosome segregation in a wide range of bacterial species. The DNA binding protein ParB (termed Spo0J in *Bacillus*

subtilis) associates specifically with origin-proximal *parS* sites and also ''spreads'' in a poorly understood way by interacting nonspecifically with adjacent chromosomal DNA. Spo0J complexes in *B. subtilis* are required for early segregation of newly replicated origins and facilitate loading of the bacterial condensin homolog SMC. using *in vitro* single-molecule imaging, we have studied the mechanism of Spo0J nucleoprotein complex formation by simultaneously observing Spo0J binding to DNA and motion of site-specific labels on the DNA chain. Our results suggest that Spo0J forms complexes by trapping long-distance loops between consensus *parS* sites and distal nonspecific segments of DNA. Detailed *in vitro* and *in vivo* analysis of mutants has allowed us to define the molecular determinants of DNA bridging by Spo0J.

3003-Pos Board B158
Acto-Myosin Contractility Rotates the Cell Nucleus
Abhishek Kumar[1], Ananyo Maitra[2], Madhuresh Sumit[1], Sriram Ramaswamy[2], Shivashankar G.V.[1].
[1]Mechanobiology Institute, Singapore, Singapore, [2]Indian Institute of Science, Bangalore, India.
The stiffest and largest organelle of the eukaryotic cell – the nucleus – is coupled to active cytoskeletal filaments. But how this organelle responds to stresses in the surrounding cytoplasm is poorly investigated. We report here the results of studies of the translational and rotational dynamics of the nuclei of single fibroblast cells, with the effects of cell migration suppressed by plating onto fibronectin-coated micro-fabricated patterns. Patterns of the same area but different shapes and/or aspect ratio were used to study the effect of cell geometry on the dynamics. On circles, squares and equilateral triangles, the nucleus undergoes persistent rotational motion, while on high-aspect-ratio rectangles of the same area it moves only back and forth. The circle and the triangle showed respectively the largest and the smallest angular speed. We rationalize our observations through a hydrodynamic approach in which the nucleus is treated as a highly viscous inclusion residing in a less viscous fluid of orientable filaments endowed with active stresses. Lowering actin contractility selectively by introducing blebbistatin at low concentrations drastically reduced the speed and persistence time of the angular motion of the nucleus. Time-lapse imaging of actin revealed a correlated hydrodynamic flow around the nucleus, with profile and magnitude consistent with the results of our theoretical approach. Coherent intracellular flows and consequent nuclear rotation thus appear to be a generic property that cells must balance by specific mechanisms in order to maintain nuclear homeostasis.

3004-Pos Board B159
Cellular Geometry Mediated Apical Stress Fibers Dynamically Couples Nucleus to Focal Adhesion
Qingsen Li, Abhishek Kumar, Shivashankar G.V.
Mechanobiology Institute, Singapore, Singapore.
Cells sense their physical microenvironment and transduce these signals through actin-nuclear links to regulate nuclear functions including gene expression. However, the spatio-temporal coupling between actin cytoskeleton and the nucleus and its modulation by cell geometry are still unclear. using micro-patterned substrates to control cell geometry, we show that perinuclear actin organization at the apical plane remodels from mesh-like structure to stress fibers. The formation of these apical stress fibers (ASF) correlated with significant reduction in nuclear height and was found to exert an active compressive load on the nucleus via direct contact with mature focal adhesion sites. We further show, using quantitative fluorescence spectroscopy experiments, that these ASFs were dynamically coupled to the nucleus via outer nuclear membrane proteins nesprin2G. Taken together, our work provides direct evidence of physical links between the nucleus and focal adhesion sites via ASFs. We suggest that such direct links may underlie nuclear mechanotransduction to regulate genomic programs.

3005-Pos Board B160
Geometric Constraints on Cells Induce Cytoplasmic to Nuclear Redistribution of Transcription Co-Factors to Regulate Gene Expression
Nikhil Jain, K. Venkatesan Iyer, Abhishek Kumar, Shivashankar G.V.
Mechano-biology Institute, Singapore, Singapore.
Physical forces in the form of substrate rigidity or geometrical constraints have been shown to alter gene expression profile and differentiation programs. However, the underlying mechanism of gene regulation by these mechanical cues is largely unknown. In this work, we use micropatterned substrates to alter cellular geometry (shape, aspect ratio and size) and study the nuclear mechanotransduction to regulate gene expression. We show that geometric constraints result in differential modulation of nuclear morphology, actomyosin contractility, histone acetylation and the activity of transcription cofactor, MRTF-A. In addition, genome-wide transcriptome analysis revealed cell geometry dependent alterations in chromosomal activity and actin dependent gene expression. Promoter analysis of these differentially regulated genes showed that serum response factor (SRF) was an essential regulatory factor sensitive to geometric cues. Further, we show that geometric constraints resulted in nuclear translocation of MRTF-A and enhanced serum response element (SRE) promoter activity. Interestingly, nuclear accumulation of MRTF-A by geometric constraints also modulated NF-κB activity. Taken together, our work provides mechanistic insights underlying the regulation of gene expression by cellular geometry.

Nucleic Acid Biophysics in vivo

3006-Pos Board B161
Multi-Scale Models of Genomic Bacterial DNA
Liang Fang, **Michael Feig**.
Michigan State University, East Lansing, MI, USA.
Detailed structural models of genomic bacterial DNA were developed using multi-scale modeling methods. Initial models were generated using a coarse-grained representation of supercoiled plectonemic DNA informed by experimental data. Conformational sampling was carried out using a Monte Carlo procedure to generate ensembles of nucleoid structures for complete genomic DNA within the constraints of known nucleoid sizes. The resulting models suggest that nucleoids are porous structures that may allow the diffusion of most proteins and protein complexes, in particular those involved in transcription and replication. The coarse-grained models were further refined with increasingly detailed representations of helical DNA up to quasi-atomistic models to serve as starting structures for realistic models of cellular environments.

3007-Pos Board B162
The Escherichia Coli Chromosome is segregated by Biased Diffusion
Nathan J. Kuwada, Paul A. Wiggins.
University of Washington, Seattle, WA, USA.
The physical mechanism responsible for accurately partitioning newly replicated Escherichia coli chromosomes into daughter cells remains a mystery. We present a quantitative characterization of the dynamical motion of the origin of replication (oriC) using a large ensemble of trajectories generated by automated complete-cell-cycle imaging. In contrast to the dynamics of chromosome segregation in eukaryotic cells, we find that the motion in this bacterium is dominated by sub-diffusive dynamics before, throughout and after the segregation process rather than processive (ballistic) motion. Instead, to maintain accurate partitioning without processive motion, we find that oriC subdiffusion is subject to a small diffusional bias (or drift velocity). Prior to oriC replication, we find that the drift velocity profile is analogous to a damped spring with equilibrium position at mid-cell. After two replicated oriC loci are distinguishable, the equilibrium position moves immediately to the quarter-cell positions and stays relatively constant for the remainder of the cell cycle, suggesting the mechanism responsible for maintaining chromosome structure may also be responsible for oriC segregation.

3008-Pos Board B163
Effect of Capsid Tail on the Ejection Dynamics of Semiflexible Polymers
Isam Hasan, Afaf Al Lawati, Muataz Al Barwani.
Sultan Qaboos University, Al Khod, Oman.
We present simulations investigating the role of the tail of a spherical viral capsid (mimicking a bacteriophage) on the ejection dynamics of a semiflexible polymer (representing viral dsDNA). We compare the ejection dynamics of a neutral polymer with that of a charged one. We find that the presence of the tail markedly slows down ejection. Our simulations suggest that this is because the last few polymer sections are trapped in the tail. Such trapping is particularly efficient for a charged polymer where the entropy of the part of the polymer outside the capsid is greatly reduced making complete ejection of the last few polymer sections difficult. Lowering the temperature further enhances this trapping.

3009-Pos Board B164
Conformational Fluctuations of Chromosomal DNA in Escherichia Coli
Rudra P. Kafle, Jens-Christian Meiners.
University of Michigan, Ann Arbor, MI, USA.
The cell is a very crowded structure, consisting of various organelles, proteins, nucleic acids, and cellular inclusions. It is the site of active, motor-driven